PULP REACTION TO ANORGANIC BOVINE DENTIN

by

MARTIN J. WALSHE

Submitted to the Faculty of the Graduate School
of Indiana University School of Dentistry in
Partial Fulfillment of the Requirements for the
Degree of Master of Science in Dentistry, 1967

ACKNOWLEDGEMENTS

The assistance of Dean Maynard K. Hine in facilitating the author's graduate dental education is gratefully acknowledged. The author would like to express his appreciation for the invaluable guidance and teaching of Dr. Ralph E. McDonald throughout the program.

The author would like to thank the faculty, secretarial staff and chair-side assistants of the Department of Pedodontics for providing an excellent clinical training in such a pleasant manner.

Dr. David F. Mitchell provided invaluable assistance in the planning and execution of this research project. For this and and many stimulating conversations the author will be forever in his debt.

The author would like to thank the remaining members of his graduate committee, Dr. Charles Gish, Dr. Richard Norman and Dr. William Shafer for their advice and support in this investigation.

Further thanks is due to Miss Shirley Shazer for preparing the histologic sections, to Mrs. Mildred Redford for her excellent care of the animals and to Dr. Denis Forest for his assistance with the micro-photography.

The author would like to thank Eli Lilly and Company for preparing the experimental material and the staff of the School of Dentistry library for their excellent service.

A special thanks is due to the author's mother, Alice Walshe and aunt, Frances Gallagher, whose love and encouragement made his professional education possible.

The past two years have been a marvelous experience and the author will always remember the kindness and hospitality of the people of the United States of America and especially of the state of Indiana.

Finally, the author wishes to thank his wife, Fionnuala, for her patience, understanding, and good humour while this course of studies was being completed.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF ILLUSTRATIONS

INTRODUCTION

The difficulty in conservatively treating exposures of the dental pulp has long been a problem in dentistry. Early observations that healing did not occur when the wound was covered by the commonly used restorations led to pulp capping with strong antiseptic and later less irritating medicaments. However, the ideal pulp capping material has yet to be found.

In order to evaluate the pulp reaction to any form of treatment, histologic analysis is essential. There should be evidence that new and experimental pulp capping materials induce repair on the normal healthy pulp before they are tested on diseased pulps in a clinical situation.

The use of an experimental animal whose dentition is closely related to that of man, allows the investigator to control measures not possible in patients and to accurately analyze the results at preferred time intervals.

Considering the effects of autogenous dentin chips on the pulp observed in earlier studies, the author felt that this aspect of pulp healing should be further examined and this study was designed to determine the pulp reaction to heterogenous dentin devoid of its antigenic potential.

REVIEW OF THE LITERATURE

Rowe[1] attributed the first description of pulp capping to Philip Pfaff, who in 1756 was the author of the first German treatise on dentistry. Pfaff was dentist to Frederick the Great, King of Prussia, and recommended capping the exposed pulp with gold plate. Investigators in the 19th Century reported various methods and techniques. Arsenic and its compounds were advocated by Spooner,[2] Arthur,[3] Chase,[4] Hitchcock,[5] and Arnold.[6] Caustics or actual cautery were recommended by Burdell[7] and Koecker.[8]

Allen[9] found greatest success with patients in good physical condition with recently exposed, non-painful pulps and advised his colleagues to use creosote on the wound followed by a pressure-relieving metallic cap.

Atkinson[10] also used creosote, but followed it with "oxy-chloride of zinc."

Christensen[11] used cobalt in his treatment of pulp exposures. However, Harlan[12] in 1893 summarized the feeling of the period when he concluded that all methods of pulp treatment varied in techniques and results, but that he, personally, favored complete extirpation.

At the turn of the century the use of formaldehyde compounds became more widespread. Investigators[13,14,15] seemed to have abandoned hopes of preserving the vitality of the exposed pulp. As a rule it was found that the pulp remained or became inflamed and that, sooner or later, necrosis occurred. In the light of his own and other workers' experiments Rebel[16] (1922) stated that "the exposed pulp is a doomed organ."

New hope for the preservation of pulp vitality arrived in the thirties with the use of calcium-containing compounds in Germany and Switzerland. Hermann's[17] publication of 1930 reporting the successful use of calcium salts, stimulated the most interest.

In 1938 Teuscher and Zander[18] reported on the action of calcium hydroxide when used in pulpotomies and described the microscopic appearance of the treated pulp including the dentinoid barrier laid down by odontoblasts near the exposure. They referred to this phenomenon as a "dentin bridge."

Zander[19] later attempted to explain the etiology of the dentin bridge by theorizing that the calcium ions in the medication would cause a precipitation of calcium salts from the blood which is normally saturated or supersaturated with calcium and phosphate ions. He also believed that the high pH of calcium hydroxide contributed to calcification since bone phosphatase was known to act best in an alkaline medium.

However, Pisanti and Sciaky[20] and Stark, et al[21] showed that the calcium was blood-borne. When radioactive calcium was used topically on the pulp, none was found in the dentin bridge whereas it was found in the bridge if injected intravenously.

Calcium hydroxide remains the most widely used pulp-capping material. Many reports in the literature[22-28] substantiate its use although most cases were evaluated on a clinical basis. Some problems are, however, associated with its use. Seltzer and

Bender[29] emphasize the fact that "...pulps may not recover completely from the effects of exposure and capping, in spite of dentin bridge formation." Asymptomatic chronic inflammation may persist for long periods of time and eventually necrosis of the pulp ensues. The presence of a bridge then complicates endodontic procedures. Seltzer and Bender mention that another undesirable effect of calcium hydroxide is the possibility of eventual complete calcification of pulp tissue in which event endodontics, if needed, often becomes an impossible procedure.

Via[30] clinically evaluated 103 cases in which pulps were capped with calcium hydroxide for an average period of 24.9 months, and considered 68.9 per cent of the cases to be unsuccessful usually due to internal resorption. Quigley[31] and Ostrom and Lyon[32] have reported failures due to pulp degeneration although dentin bridges had been built.

Antibiotics and their compounds have attracted considerable attention from dental investigators in recent years. In 1942, penicillin therapy first appeared in the Quarterly Cumulative Index Medicus,[33] and it was only a matter of time until the drug was used as a pulp capping agent.

However, there is a paucity of well controlled, thoroughly documented investigations on the pulp reaction to antibiotics. Bower[34] and Kutscher[35] in separate studies reported encouraging results using penicillin as a capping agent. Kutscher capped cariously exposed pulps and found a 98 per cent success rate

after six weeks. Other investigations showed a similar pattern of success.[36,37,38]

Seltzer and Bender,[39] and Burke and Holmes[40] did not find quite the same degree of repair. Seltzer and Bender in fact observed that pulpal necrosis occurred in every instance when an aqueous solution of 250,000 units of penicillin was used on vital pulp tissue of dogs. More recent results, however, are more encouraging.[41]

Antibiotics have been combined with various medicaments, but the techniques used and the results found have been equally varied.

The use of corticosteroids with antibiotics deserves some mention. The theory that an antibiotic would destroy the bacteria and the corticosteroid would reduce the inflammation was a laudable one, but the results of investigations so far have not been very encouraging. Clinically, the treatment seemed to be highly successful,[42-46] but the histological appearance of pulps treated was quite different. Baume[47] summarized the histologic findings following pulp capping with corticoid-containing medicaments. He noted metaplastic changes of the pulp tissue, irreversible inhibition of dentine formation and chronic inflammation which caused death without symptoms later. The absence of dentinogenesis was noted and thought to be due to the suppression of RNA formation and protein synthesis by the corticosteroid.

However, Lawson and Mitchell,[48] in a double blind study capped 52 teeth with demonstrated presence of painful pulpitis, and evaluated the results up to 164 days. There were no failures among the experimental group. In a limited histologic analysis the experimental compound showed a distinct advantage over the starch control. Mullaney, et al,[49] in continuation of the same study re-examined a portion of the original sample and found that of 21 treated teeth 15 were still considered to be successful. Although this represented a considerable drop from the 100 per cent success in the preliminary investigation, it appeared that painful pulpitis was a reversible process and that the corticoid-antibiotic mixture is of therapeutic value.

The confusing reports relating to the use of corticosteroid-antibiotic pulp capping agents continue. In a recent Journal two separate studies are reported in which the same medicament was used but totally different results were found. Olsen[50] found success, but Fiore-Donno and Baume[51] found chronic inflammation.

The role bacteria play in the healing of pulp exposures was best illustrated by Kakehasi, et al,[52] in 1865. They found that in a germ-free environment the exposed pulps of rats' teeth healed even though the cavities were not restored and the pulps were exposed to food throughout the experiment. The control teeth of normal animals on a normal diet were invariably necrotic. It was noted that the bridging of pulps in the experimental animals was considerably aided by the presence of dentin chips

which had been accidentally introduced during cavity preparation. The chips seemed to act as centers of reparative dentinogenesis.

The Role of the Dentin Chips

In 1879 Stellwagen[53] urged his colleagues to use the "natural dentin for capping the exposed teeth pulps." He advised against the use of medicaments such as creosote, carbolic acid, chloride of zinc and alcohol, which were popular at the time, maintaining that these drugs incited a foreign body reaction. He suggested that the operator "...allow the wound to glaze over and then with a clean, sharp excavator shave off from the walls of the cavity enough healthy dentine to cover over the point to be capped." He theorized that the dentin chips would aid in pulp healing, as skin grafts did in lesions of the skin, and he claimed the highest successes with his technique over a period of 18 months.

Later, Buckley[54] invented his artificial dentin hoping it would succeed on the same principle. He tried to include calcium phosphate in the same percentage as found in natural dentin. His preparation, "Dentinoid," consisted of: Calcium Phosphate 60 per cent, Thymol 2 per cent, Thymol Iodide 3 per cent, Bismuth Subritrate 5 per cent and Resin 10 per cent. The medicaments were included to sterilize the dentin and to stimulate the odontoblasts towards dentinogenesis.

Later, several other investigators reported on the use of dentin chips. Datwyler,[55] Kronfeld,[56] Feldman,[57,58] and Hellner[59]

noted that dentin chips were imbedded in "callus" or "osteoid tissue" in the pulp and thought that the fragments aided in the healing process.

There is considerable variation in the results of pulps treated with dentin chips. (This may be due to variation in the selection of teeth). Neuwirt[60,61] and Pribyl[62] examined treated pulps clinically and histologically and reported all cases successful. However, Zajfe and Schatzker[63] and Loewenstein[64] found that the majority of their cases failed.

One of the better studies of the pre-war era was conducted in Switzerland by Hoffman.[65] Working on healthy teeth which were scheduled for extraction, Frau Hoffman capped the exposed pulps with autogenous and homogenous dentin. The homogenous dentin was collected from extracted teeth by drilling and later was boiled. Autogenous chips were drilled from the walls of the cavity and immediately placed over the exposure. They in turn were covered by a silver plate. At three to nine month intervals, 20 of 34 teeth treated with autogenous chips were examined histologically and showed complete closure of the exposure. Five showed complete closure, plus infiltration, and six failed due to infection. Of the 18 teeth capped with homogenous dentin 13 were available for histologic examination, only four showed full closure, four others had closed but had a lymphocytic infiltration and three were abscessed. Two teeth were lost due to poor fixation.

Further reports in the literature relating to the pulp reaction to dentin chips were made by Castagnola,[66] Glass and Zander,[67] Van Huysen and Boyd,[68] Kalnins and Frisbie,[69] Doyle,[70] Spedding,[71] and Baker.[41] All studies were complicated by the fact that other pulp capping materials were actually being tested and a fair assessment of the chips was difficult.

The use of dentin chips has not been confined to pulp-capping, Schaffer[42] used cementum and dentin fragments to induce osteogenesis in artificially created infra-bony pockets in the dog and monkey. Autogenous fragments proved osteogenic in the dog and autoclaved human tooth fragments showed some success in the monkey. He noted the absence of a foreign body type reaction. Kuttler,[73] when proposing a root canal filling technique, recommended the use of the clean autogenous dentin filings from the canal to bring about apical repair in a shorter period of time. He noted the formation of osteoid material around the implanted chips at the tooth apex.

STATEMENT OF PROBLEM

A review of the literature shows that superficial lesions of the dental pulp continue to present a problem to the operative dentist. Calcium hydroxide, the most thoroughly investigated medicament, shows some unfavorable reactions, even when applied to normal vital pulps in ideal situations. The search for a more successful medicament must continue and experimental materials must prove to be acceptable and should induce repair of the normal pulp before they can possibly be tried on the diseased pulp.

It has been noted that autogenous dentin chips, accidentally introduced to the pulp, seem to act as "centers of dentinogenesis" and play a major role in the bridging of the exposure. Dentin chips apparently do not cause any significant inflammation and were well tolerated by the pulp tissue.

This study was designed to determine if heterogenous dentin chips, devoid of their antigenic potential, would have a similar effect on the pulp.

PREPARATION OF THE MATERIAL

It was decided to use the lower incisors of the young calf as the source of bovine dentin. There is a possibility that the extract may be more potent than if taken from an older animal[74] and also it was thought that the extractions would be less difficult. The calf mandibles were obtained at a local abattoir and the teeth were removed with the aid of a scalpel and surgical forceps #101. The crowns of the teeth were removed from the root with the aid of a diamond disk after which the radicular pulps were removed. The remaining roots were trimmed of all soft tissue, stored in normal saline and sent to the laboratory for processing.

The remaining laboratory work was carried out by a pharmaceutical company.* The purpose of the procedure was to degrade antigenic proteins and was the procedure used by Prudden and Allen[75] in preparing a bovine cartilage extract to produce an acceleration of wound healing. They provide a thorough review of the use of their technique and its clinical results in their article.

The procedure of acid-pepsin hydrolysis† of bovine dentin is summarized as follows:

1. Suspend 10 G. calves teeth roots in 80 ml. 0.6 per cent acetic acid containing 300 mg. pepsin, N.F. Stir this mixture at 70° for five hours. Then wash the teeth with hot tap water and deionized water and dry overnight in a vacuum desiccator. Recovery: 8.7 G.

---

* Eli Lilly & Co., Indianapolis, Indiana. Experiment No.
† Belgian patent 650626 assigned to L.L. Balassa.

2. Grind the deproteinized teeth in a hand meat grinder. Slurry the small chips in n-hexane to effect defatting. Dry overnight in a vacuum desiccator.

   Recovery: 4.90 G. (Most of this loss occurred during the grinding operation. It was difficult to keep tooth chips from flying out of the grinder.)

3. Grind the deproteinized, defatted tooth chips in a rotary ball mill for 14 hours. Dry sieve the resulting powder, collecting the powder that passed through a 300-mesh screen.

   Recovery: 2.30 G.

4. Sterilize by autoclave

The final powder was white and extremely fine in texture. In order to facilitate the application of the powder to the small exposures of the monkey teeth, it was decided to mix it with some suitable liquid. Test mixes with distilled water and normal saline proved to be inadequate. However, when mixed with 1 per cent methyl cellulose, the handling properties were somewhat improved.

EXPERIMENTAL PROCEDURE

Two 10-pound female Macaca Speciosa were utilized in this study because of the encouraging reports from Kling and Orbach[76] and Baker.[41]

The animals were procured from a distributing center in Detroit where quarantine restrictions had already been observed. Therefore, only three days was lost before the experiment began. Each animal was, however, given a routine dusting with five per cent DDT powder and a gross physical examination at the time of arrival, to insure against changes in condition in transit.

The animal was removed from its cage and handled in general as described so thoroughly by Spedding.[71] The dosage of anesthetic was judged primarily by weight - each animal receiving 5 ml. (25 mg. per ml.) Numbutal* intravenously. While awaiting the onset of anesthesia, the animal was kept in a small handling cage to prevent accidents and 1/2 ml. atropine sulphate† (1/150 gr. per c.c.) was administered to reduce secretions.

Each animal was intubated and an intravenous drip of I. V. Ringers Lactate Solution§ was mounted. A pack was placed in the oro-pharynx, the head was supported on a pillow of paper towels.

---

* Veterinary Menbutal Sodium, Abbott Laboratories, North Chicago, Ill.
† Atropine Sulphate, Eli Lilly, Indianapolis, Ind.
§ I. V. Ringer's Lactate Solution, Abbott Laboratories, North Chicago, Ill.

Body heat was maintained by placing a piece of wrapping paper over the torso and the animal was ready for the operation. Preoperative radiographs were taken to ascertain pulp size and position.

The teeth were cleaned of all visible debris and wiped with a cotton gauze soaked in 70 per cent alcohol. Due to the total lack of saliva, the rubber-dam was not considered necessary and was not used.

Experimental Teeth:

Black Class V cavities were prepared with an air-cooled, high speed No. 57 burr. The floor of the cavity was deepened with a No. 3 round burr rotating at slow speeds until the shadow outline of the pulp was noticeable. All debris was removed from the cavity and the floor was punctured with a jackette scaler until hemorrhage was produced. This procedure was adopted to prevent mutilation of the pulp tissue by the burr and by numerous autogenous chips, thus facilitating a more accurate histological diagnosis later. The technique is extremely slow and painstaking, but only four teeth in the entire experiment were exposed accidentally by the burr.

After the hemorrhage was arrested with sterile cotton pellets, the anorganic bovine dentine powder was mixed with methyl cellulose and applied to the exposure site. Three teeth were exposed at the same time to save numerous mixings of the

powder. Sufficient amounts of the capping material were placed, in order to prevent any of the sealer coming in contact with the pulp.

The cavities were finally sealed with a zinc oxide-eugenol cement.*

In each animal, all molars, premolars and the maxillary central and lateral incisors were utilized as experimental teeth.

Control Teeth:

Two types of controls were utilized.

(a) Autogenous Dentin Chips: Since the experiment was designed to study the pulp reaction to bovine dentin, the monkey's own dentin chips were used as one control measure. The four cuspids of each animal were utilized for this purpose. Class V cavities were prepared as described previously, but after the exposure hemorrhage was arrested, dentin chips were drilled from the walls of the cavity and packed over the wound. When the wound was completely covered, zinc oxide-eugenol was used to seal the cavity.

(b) Calcium Hydroxide: Calcium hydroxide, second control, was chosen because its action on the pulp is now well documented

---

* Temrex Cement, Interstate Dental Co., Inc., New York, N.Y.

and because of its widespread and popular use as a pulp-capping agent. The lower incisors of each animal were capped in this manner, the technique being the same as described previously, except for the final medicament.

Post-Operative Care

All restorations were checked for smoothness, in order to avoid soft tissue irritation. The mouth was washed out with water and the pack and tube were removed. The animal was observed closely during the recovery period as respiratory failure sometimes occurs then. The maintenance of body heat was considered essential during this period and an analgesic* was mixed with the diet in order to relieve any post-operative discomfort.

Until the day of sacrifice, the condition of the stools, body weight and behavior were kept under surveillance.

Surgical Procedures

The first animal was sacrificed after 21 days, the second after 42 days.

The animal was anesthetized as described previously and preoperative radiographs were taken. The teeth were removed with the aid of surgical forceps No. 101. Occasionally, it was necessary to remove the buccal plate of bone and the automatic bone impactor (Dudley) was used.

---

* Darvon Compound, Eli Lilly Co., Indianapolis, Ind.

As the teeth were extracted, they were placed in separate pre-labeled bottles containing 10 per cent formalin solution for primary fixation.

Laboratory Procedure

The teeth were ground on the mesial or distal surface with a rotating stone wheel under a water spray until the outline of the pulp could be seen. The teeth were then returned to their bottles for complete fixation of the pulp.

Decalcification and the remaining laboratory work was performed by a laboratory technician. Serial paraffin sections seven microns thick were made through the exposure site and stained with hematoxylin and eosin in the conventional manner.

The teeth of the second (42 days) animal were also examined for the presence of bacteria using the Brown and Brenn stain.[77] The reason for this procedure will be explained later.

RESULTS

The first animal was sacrificed after 21 days, the second after 42 days. At each day of sacrifice a thorough clinical and radiographic examination was performed prior to the extraction of the teeth.

Clinical Evaluation

Observations at 21 days

A thorough clinical examination was performed on the animal to determine tooth mobility, condition of the restoration and the normalcy of the supporting tissues. Two of the restorations seemed to have deteriorated. There was no evidence of change in the supporting tissues.

Observations at 42 days

A similar examination was performed. No evidence of pulp pathosis was found. Three of the restorations were displaced.

Radiographic Evaluation

No radiographic evidence of pulp pathosis was found at either the 21 or 42 day observation period.

Histologic Evaluation

Histologic Evaluation 21 days after capping. (Table II)

Of the 17 permanent tooth pulps capped with anorganic bovine dentin, seven appeared normal and were depositing reparative dentin to repair the exposure.

The almost total absence of any inflammation was unique. Generally speaking, the exposures were almost closed completely by reparative dentin of a non-tubular variety. The reparative

dentin was deposited not only on the wound surface adjacent to the capping material, but also around any of the material which was deep in the pulp tissue.

A similar reaction occurred adjacent to autogenous dentin chips which were accidently introduced into the pulp. Indeed, these dentin chips appeared to act as centers of dentinogenesis and the reparative dentin of one seemed to coalesce with the next until the bridge was complete.

The reparative dentin was paler staining and as mentioned earlier of the non-tubular variety. The odontoblasts adjacent were not aligned in the normal fashion, but were unevenly distributed. As a group, the seven pulps were considered an unquestionable success.

The ten other pulps of the animal did not react in such a favorable manner, although the pulps of two teeth might be considered partially healed. One of these was the lower left first bicuspid which was severely traumatized by the burr. In this instance, the pulp showed a severe local inflammation and some spaces which might possibly have been minute abscesses or perhaps were merely artifacts. Moreover, reparative dentin production was retarded and the exposure site was still open.

The majority of the failures showed evidence of abscess formation and local, severe inflammatory cell infiltration. Of the ten pulps, two showed an acute inflammatory response of the pulp throughout the chamber and canals, but in the remaining

eight, the inflammatory infiltrate was localized and confined to the immediate area of the exposure.

Two of the four teeth capped with autogenous dentin chips, were free of inflammation and showed abundant reparative dentin closing the exposure. Of the two remaining teeth, one showed acute inflammation locally but an otherwise healthy pulp while the other tooth showed diffuse inflammation with some necrosis. The reparative dentin in all these pulps was the same type as that produced in pulps capped with bovine dentin.

The teeth capped with calcium hydroxide showed classical reactions of surface necrosis, followed by deposits of reparative dentin with a mild hyperemia of the adjacent tissue. The repairing dentin seemed to have been deposited in a more urgent fashion since cells were frequently observed enclosed in the mass.

Histologic evaluation 42 days after capping (Table III)

Due to the unusual distribution of the failures in the first monkey, it was decided to keep a section from each block and stain for bacteria with the Brown and Brenn technique in the teeth of the second animal.

The distinction between success and failure was not as definite as in the 21-day analysis. Of the 20 teeth capped with the experimental bovine material, seven were considered successful, nine were considered failures and four were "border-line."

The seven successful pulp-cappings showed normal healthy pulps and bridging at the exposure site, which was either complete or almost complete.

Two of the nine failures showed total necrosis of the pulp, but the majority showed abscess formation in the chambers. No reparative dentin was found in any of these pulps.

The "border-line" pulps showed mild inflammation locally and some limited reparative dentin formation. These pulps showed reparative dentin formation around dentin chips and the capping material which had accidentally been pushed deep into the canal. It appeared that repair had commenced but had ceased before the task was complete. In all these teeth, the inflammation was very localized, the remainder of the pulp being quite healthy.

The four pulps capped with autogenous chips were normal and healthy and the exposures were walled off by reparative dentin. The four lower incisors capped with calcium hydroxide were also considered successful.

When the sections stained with hemotoxylin and eosin were compared with those stained for bacteria, a direct correlation was found between the presence of bacteria and lack of reparative dentin. Bacteria were also found in or adjacent to those pulps which had mild reactions, but showed limited calcific repair. No bacteria were found in the pulps which healed successfully.

In one of the successfully repaired teeth, bacteria were found in the cavity.

TABLES AND FIGURES

TABLE I

Histologic Results for Pulps of Control Teeth

Tooth	Exposure Size	Capping Material	Inflammatory Status of Pulp	Reparative Dentin Formation	Pulpal Reaction
Animal Number I --- 21 Days					
LR3	Small	A.D.C.	Severe	Neg.	2
LL3	Medium	A.D.C.	Severe	Neg.	3
UR3	Small	A.D.C.	Mild	Marked	1
UL3	Small	A.D.C.	Mild	Marked	1
LR1	Medium	$Ca(OH)_2$	Mild	Marked	1
LR2	Medium	$Ca(OH)_2$	Mild	Marked	1
LL1	Small	$Ca(OH)_2$	Mild	Marked	1
LL2	Medium	$Ca(OH)_2$	Mild	Marked	1
Animal Number II --- 42 Days					
LR3	Small	A.D.C.	Mild	Marked	1
LL3	Medium	A.D.C.	Mild	Marked	1
UR3	Small	A.D.C.	Mild	Marked	1
UL3	Small	A.D.C.	Mild	Marked	1
LR1	Small	$Ca(OH)_2$	Mild	Marked	1
LR2	Small	$Ca(OH)_2$	Mild	Marked	1
LL1	Small	$Ca(OH)_2$	Mild	Marked	1
LL2	Small	$Ca(OH)_2$	Mild	Marked	1

Abbreviations

A.D.C. --- Autogenous Dentin Chips

Symbols

1 --- Satisfactory
2 --- Unsatisfactory (inflammation)
3 --- Unsatisfactory (necrosis)

TABLE II

Histologic Results of Pulps Capped with Anorganic Bovine Dentin

Tooth	Exposure Size	Inflammatory Status of Pulp	Reparative Dentin Formation	Pulpal Reaction
Animal Number I --- 21 Days H and E Stain				
UR1	Small	Severe	Minimal	2
UR2	Large	Severe	Neg.	2
UR4	Large	Severe	Neg.	2
UR5	Large	Severe	Neg.	2
UR6	Small	Severe	Neg.	2
UR7	Small	Severe	Neg.	2
LR6	Large	Severe	Neg.	3
LR5	Small	Severe	Neg.	2
LR4	Small	Severe	Minimal	2
LL4	Large	Severe	Minimal	2
UL1	Small	Mild	Marked	1
UL2	Small	Mild	Marked	1
UL4	Small	Mild	Marked	1
UL5	Medium	Mild	Marked	1
UL6	Medium	Mild	Marked	1
LL5	Small	Mild	Marked	1
LL6	Small	Mild	Marked	1

Symbols
1 --- Satisfactory
2 --- Unsatisfactory (inflammation)
3 --- Unsatisfactory (necrosis)

TABLE III

Histologic Results of Pulps Capped with Anorganic Bovine Dentin

Tooth	Exposure Size	Inflammatory Status of Pulp	Reparative Dentin Formation	Pulpal Reaction	Locus of Bacteria*
Animal Number II --- 42 Days			Bacterial Correlation*		
UR2	Small	Severe	Neg.	3	CP
UR4	Small	Moderate	Minimal	2	i
UR5	Small	Severe	Neg.	2	i
UR6	Large	Severe	Minimal	3	P
UR7	Small	Severe	Neg.	2	P
LR4	Small	Severe	Neg.	2	P
LR5	Small	Mild	Moderate	2	C
LR6	Small	Severe	Minimal	2	CP
LR7	Small	Severe	Neg.	2	C
UL6	Medium	Severe	Neg.	3	P
UL7	Small	Moderate	Minimal	2	CP
UL2	Small	Severe	Neg.	3	P
UL4	Medium	Mild	Marked	1	C
UL5	Small	Mild	Marked	1	0
LL4	Small	Mild	Marked	1	0
LL5	Small	Moderate	Moderate	2	C
LL6	Small	Mild	Marked	1	0
LL7	Small	Mild	Marked	1	0
UL1	Small	Mild	Marked	1	0
UR1	Small	Mild	Marked	1	0

Abbreviations
0 - No bacteria found
C - Bacteria in cavity
P - Bacteria in pulp tissue
i - Bacteria ingested by phagocytes

Symbols
1 - Satisfactory
2 - Unsatisfactory (inflammation)
3 - Unsatisfactory (necrosis)

* Brown and Brenn, special bacterial stain

Figure 1. The anesthetised animal, showing intubation tube, tongue maintainer, and the intravenous drip.

Figure 2. Cuspid exposure. The incisors and the second bicuspid are capped and sealed.

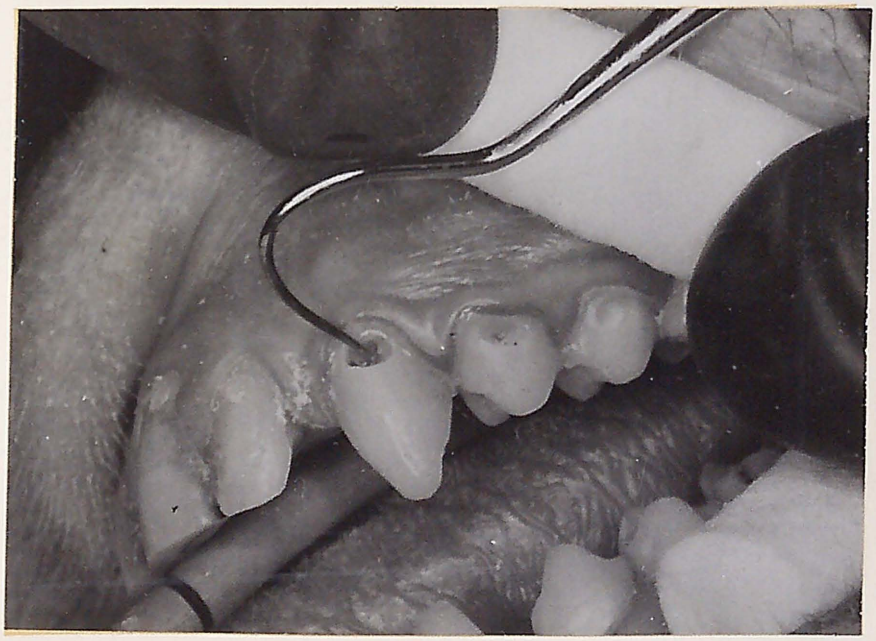
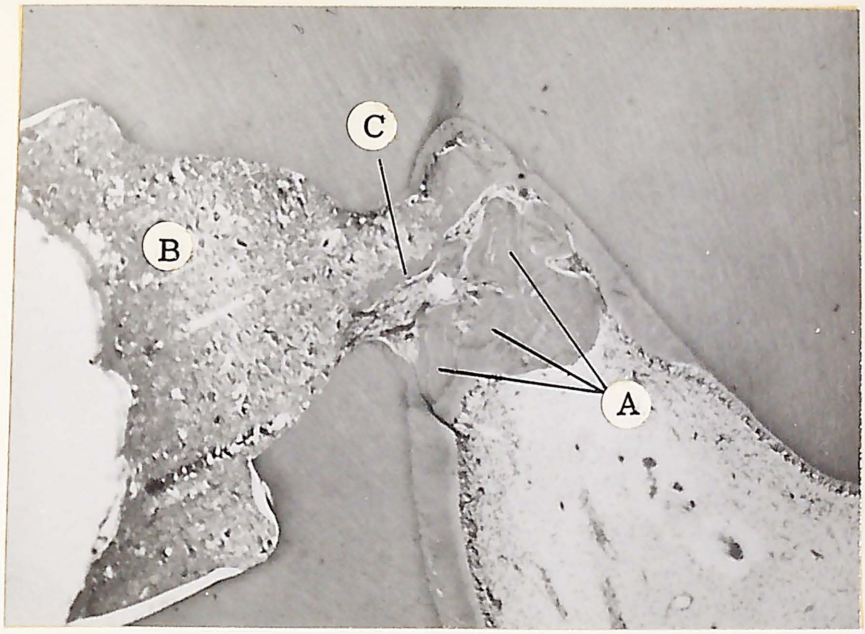


Figure 3. Permanent tooth capped with bovine dentin. (21 days).
A. Autogenous dentin chips
B. Bovine dentin material
C. Calcific bridge laid down by the pulp
Hematoxylin and eosin stain.
Original magnification X50

Figure 4. Same section as above at high power.
Original magnification X75


C
B
A

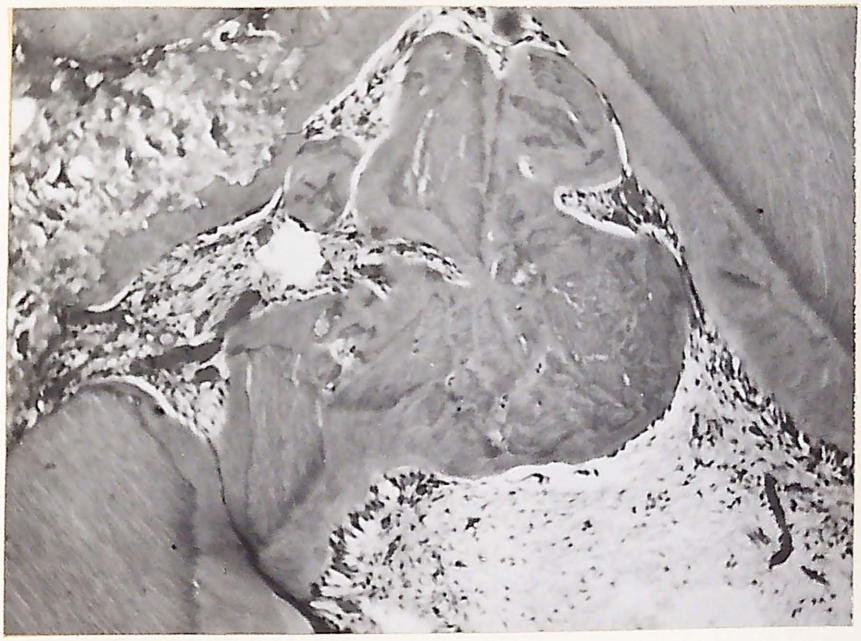


Figure 5. Pulp capped with bovine dentin (42 days). Note reparative dentin formation at the mouth of the exposure and also around the particles free in the pulp.
A. Anorganic bovine dentin
B. Bridging repair of exposure
C. Cavity
Hematoxylin and eosin stain.
Original magnification X75

Figure 6. Control tooth, capped with autogenous dentin chips only, showing reparative dentin formation and almost complete closure of the wound (21 days).
Hematoxylin and eosin stain.
Original magnification X50

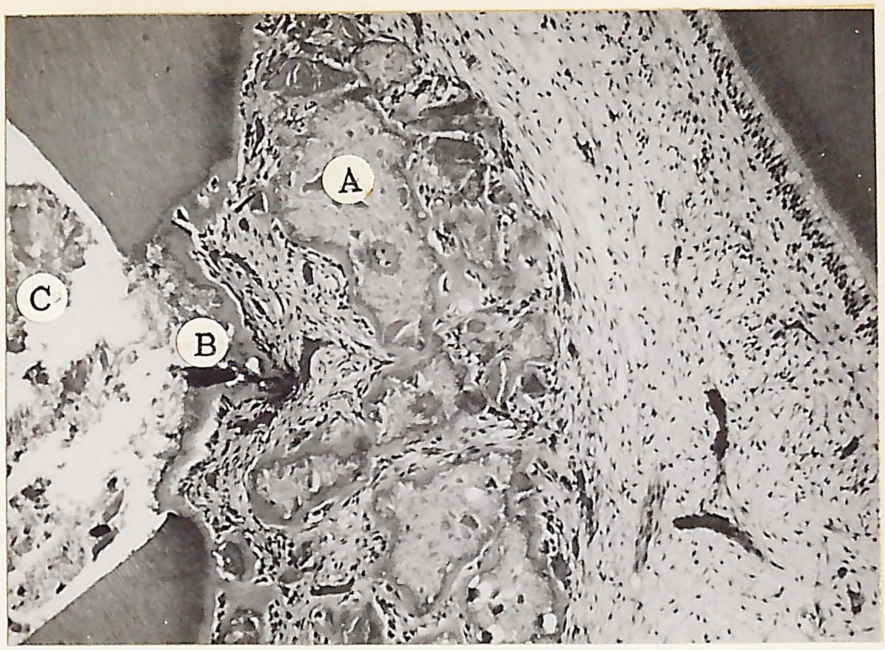

A
B
C

Figure 7. Pulp capped with bovine dentin (42 days). Total necrosis.
Hematoxylin and eosin stain.
Original magnification X75

Figure 8. Same specimen stained by Brown and Brenn technique.
Note presence of bacteria in the mouth of the exposure.
Brown and Brenn stain.
Original magnification X75

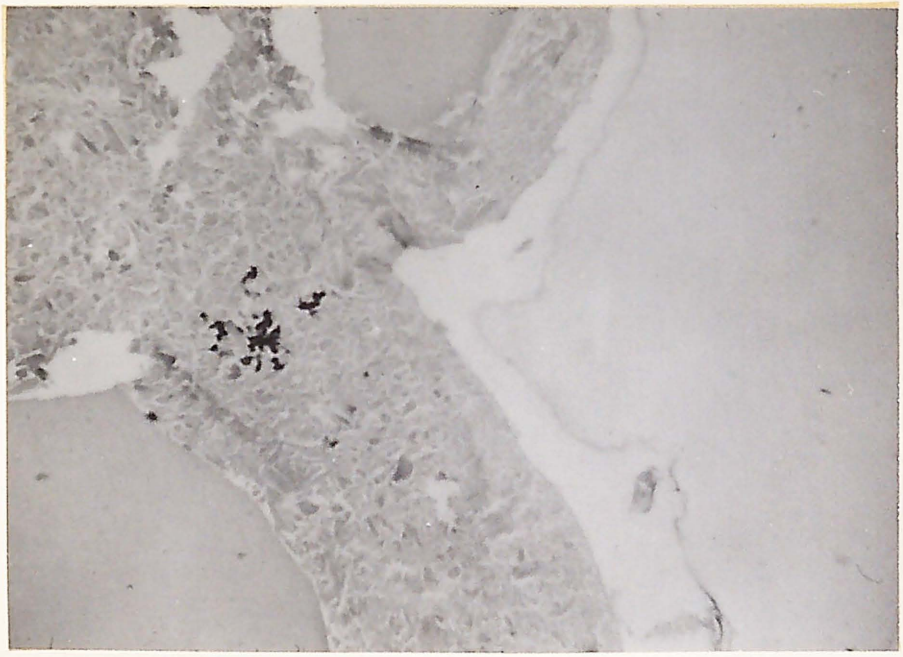
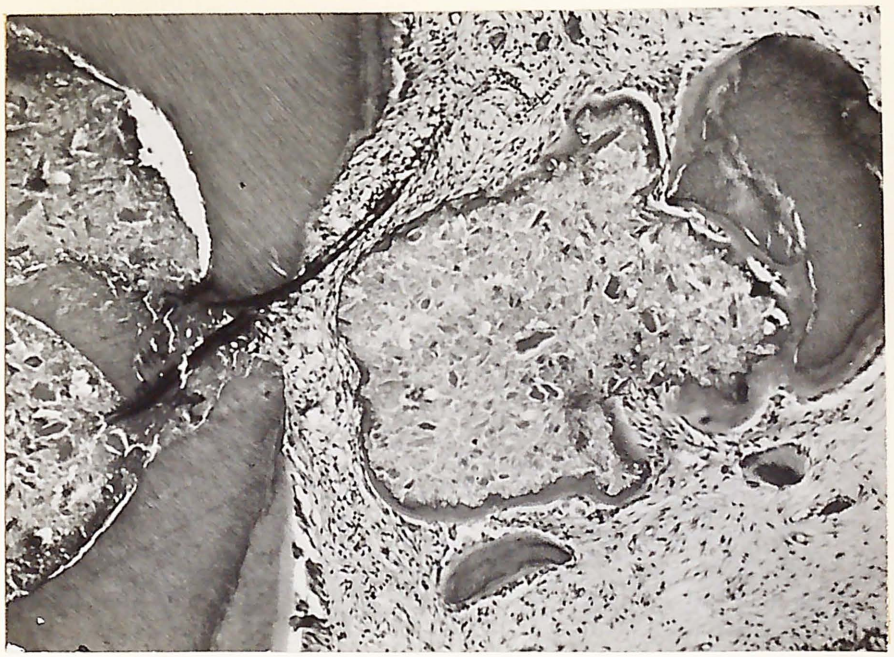
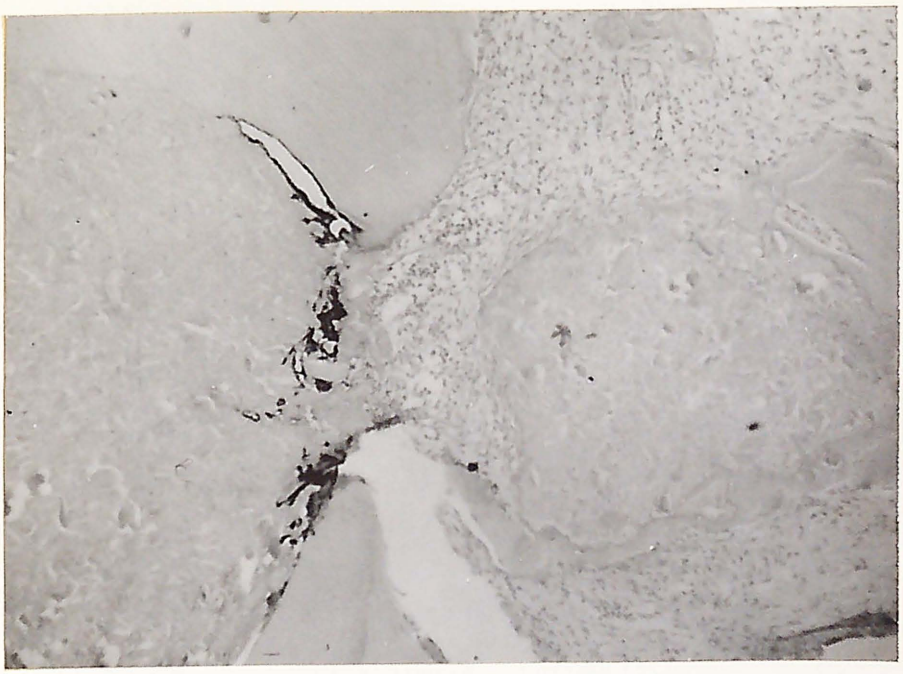


Figure 9. Tooth capped with anorganic bovine dentin (42 days). Note lack of reparative dentin adjacent to exposure in comparison to deeper in the pulp. Hematoxylin and eosin stain. Original magnification X75

Figure 10. Same tooth stained for bacteria. Compare locale of bacteria with distribution of calcific repair in Figure 9. Brown and Brenn stain. Original magnification X75

Figure 11. High power view under oil of area showing bacteria in cavity interface and in the dentin tubules. Brown and Brenn stain. Original magnification X100





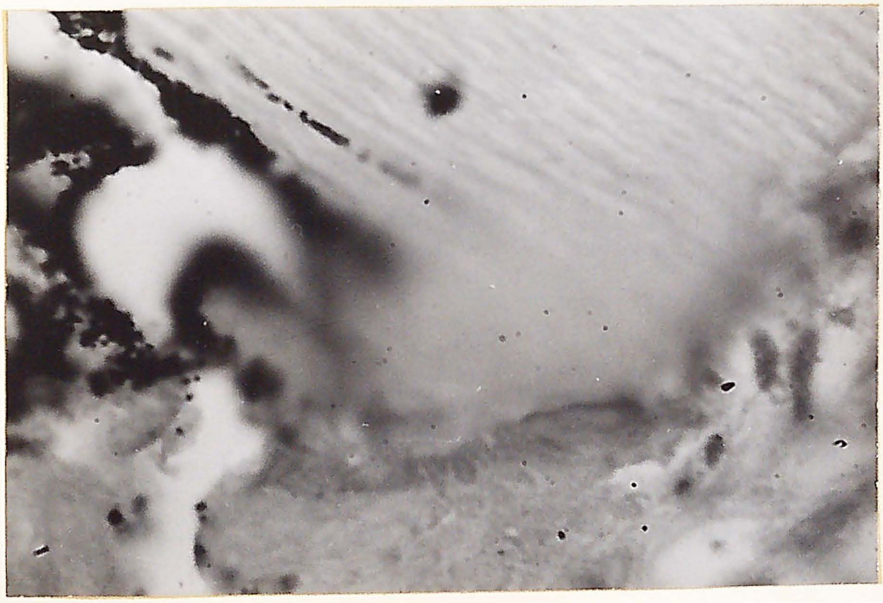
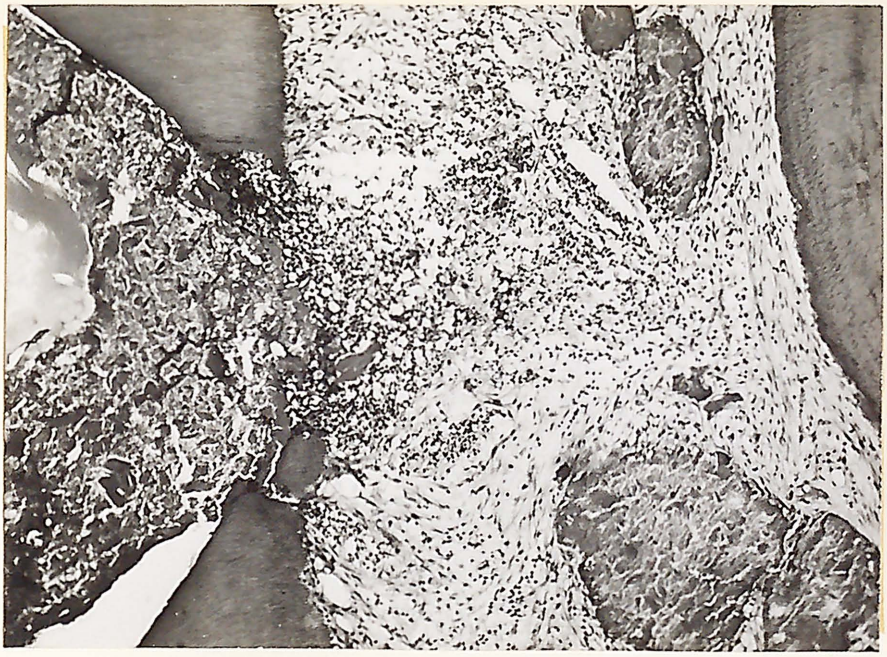
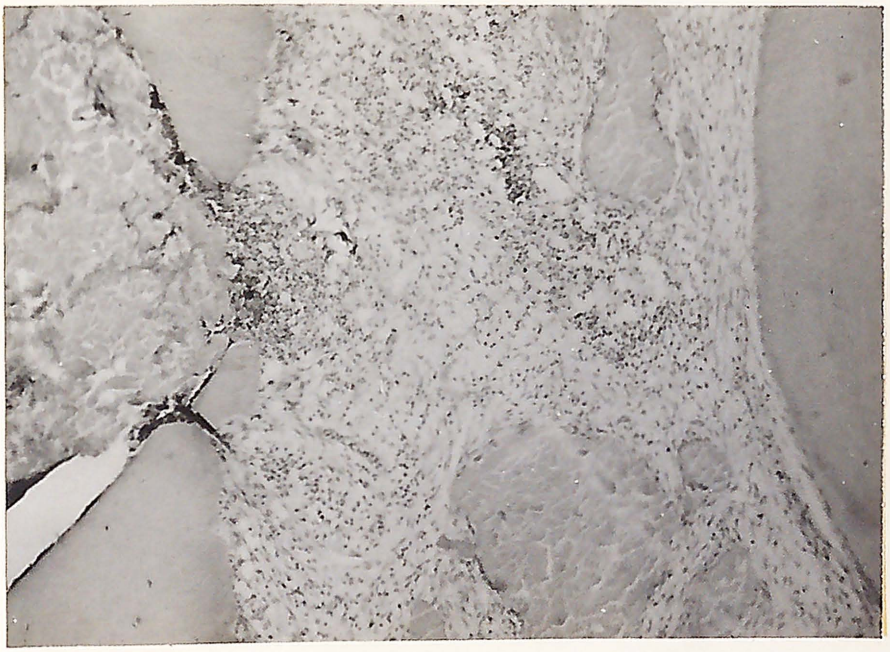


Figure 12. Tooth capped with anorganic bovine dentin (42 days). Note severe inflammation and absence of calcific repair.
Hematoxylin and eosin stain.
Original magnification X75

Figure 13. Same tooth stained with Brown and Brenn technique. Note bacteria in the cavity and pulp.
Brown and Brenn stain.
Original magnification X75





DISCUSSION

When the histological sections of the first (21 day) monkey were examined, it was noted that the majority of teeth of the left side of the mouth demonstrated evidence of favorable response to the capping material, while those on the right side did not. This unusual distribution suggested that the failures were due to contamination of the pulps by bacteria, rather than due to a specific action of the anorganic bovine dentin. The presence of a normal pulp in one tooth and abscess production in another added to the assumption and it was decided to stain one section of each pulp of the second animal, using the Brown and Brenn[77] technique. Thus, it was possible to examine the exposure area of each pulp, using both the routine (H and E) stain and the bacteriologic stain and compare the results.

The results, as shown in Table III, are interesting. Bacteria were found in all the severely inflamed pulps and also in those pulps where the inflammation was mild, but the repair was limited (Figure 9). Obviously, the role of bacteria in pulp healing is a major one. Kakehasi, et al,[52] have unquestionably shown this. Therefore, it seems logical that future studies relating to pulp healing should be conducted with the most aseptic procedure possible and all pulps should be stained for bacteria, in order to help the investigator decide whether inflammation is due to a specific effect of the medicament, or due to contamination by bacteria. It is possible that a promising pulp capping material

might be condemned as phlogogenic, whereas in actual fact the poor results might be due to bacterial infection.

It is regrettable that the Brown and Brenn technique was not used on the pulps of the first animal, but the author did not expect contamination to be a factor in the results. However, it seems reasonable to assume that bacteria were the cause of the failures.

The question of how bacteria gained access to the pulps is difficult to answer, since every attempt was made to follow an aseptic procedure during operations. Saliva was not a problem, but perhaps a separate sterile burr and explorer should have been used for each tooth. It is also possible that the sealer restoration of zinc-oxide and eugenol leaked postoperatively, but the material was chosen because of its superior sealing properties.[78] These properties could have been altered by poor mixing, yet all mixing was done by an experienced dental assistant. It did appear on some sections that the bacteria had leaked into the pulp by way of the cavity interface (Figure 13). Whether this was due to an inferior restoration or due to manipulation of the restorations by the animal is open to dispute. At any rate, it appears that future investigators would be advised to fill the cavities with not only the sealer, but also with a coat of varnish and an outside covering of amalgam. In clinical practice, capped pulps are often covered by a temporary restoration during the

observation period. The results of this study seem to indicate that this procedure may not be advisable and that a more formidable restoration is needed.

In this study, a relationship appeared to exist between the degree of pulp inflammation and the amount of calcific repair. Pulps severely inflamed did not initiate bridging, while the greatest amounts of reparative dentin were observed in pulps which had little or no inflammation. This relationship has been observed by other investigators[38,41,52] and the point is well illustrated in Figures 9 and 10. Reparative dentin is abundant deep in the pulp, but quite limited near the exposure where the bacteria are present.

Most pulps capped with autogenous dentin chips were successfully repaired. The chips seemed to stimulate islands of reparative dentin which joined to form the bridge. No evidence was found of the delayed healing and inflammation as described by Kalnins and Frisbie.[69] It is possible that the inflammation they described was due to contamination of the pulps by bacteria. The healing action of dentin chips is probably the reason why numerous undiagnosed traumatic exposures heal uneventfully in dental practice.

In spite of a conscious effort to prevent it, most of the experimental pulps included autogenous dentin chips. However, in no instance were the chips so numerous that a fair assessment

of the experimental material was impossible. Anorganic bovine dentin proved to be dentinogenic and non-inflammatory in the absence of bacteria.

Subramanian,[79] in 1961, did not achieve calcific repair with anorganic bone (autoclaved), but Ostrom and Lyon,[32] in 1962, found favorable repair using bone treated with ethylenediamine. It is difficult to reach a conclusion on this evidence, but the method of treating the material for capping may be important. It is equally difficult to determine whether bone or dentin has the greater potential as a pulp-capping agent.

Complete bridging, as determined by serial sections, was not observed in all of the pulps treated with the bovine dentin. Bridging, when present, was usually incomplete in one small area approximately 20 microns wide on the average (Figure 4). It was noted that cells from the pulp tended to invade the cavity and their presence may have prevented bridging. A somewhat similar situation is seen in pulps which appear to be totally calcified, but on histologic examination a thin strip of pulp tissue is found. The cauterizing action of calcium hydroxide may account for the fact that this phenomenon was not observed with the calcium hydroxide controls. The high pH of the material would kill any cells in the immediate area. Indeed, the cauterizing properties of calcium hydroxide may be the reason for its high rate of success. Not only is the bridging usually complete, but the high pH may also kill bacteria in the immediate area of the pulp.[63]

No significant difference was found between the types of reparative dentin induced by either the autogenous or heterogenous dentin. The reparative dentin was acellular and of the non-tubular variety. The calcium hydroxide, however, generally induced an osteo-dentin. It is difficult to say whether either type is more desirable.

As always, more research is needed. The fact that anorganic bovine dentin is dentinogenic in the absence of bacteria warrants further study. Probably a more potent extract of dentin exists and a study should be designed to screen the various extracts. Seltzer, et al,[81] tried to induce calcific repair in exposed pulps with alkaline phosphatase. Reparative dentinogenesis occurred in 66 per cent of the experimental teeth, but they felt that the evidence was not conclusive and recommended further investigations. Johanson, et al,[82] capped rat molar pulps with collagen and chondroitin sulphate, hoping that the materials would provide a ready-made matrix for calcification. Their hypothesis did not hold true, but they considered the formation of the matrix was the most important aspect of bridge building and suggested that further research be done in this area.

The perfect pulp capping material will probably be a biologic extract of bone or dentin. Perhaps a universal healing agent common to all tissue will eventually be found but before then, a lot of thoroughly documented, well disciplined research is needed.

SUMMARY AND CONCLUSIONS

Previous investigations have suggested that autogenous dentin chips, accidentally introduced into the pulp, initiate reparative dentinogenesis and play a significant role in bridging the exposure area. This study was designed to determine if bovine dentin, devoid of its antigenic potential, would have a similar effect on the dental pulp.

The pulps of 37 permanent teeth of two monkeys were surgically exposed and capped with bovine dentin, which had been subjected to acid-pepsin hydrolysis. Eight pulps were capped with autogenous dentin chips from the cavity walls and another eight pulps were capped with calcium hydroxide, making a total of sixteen control teeth. All cavities were sealed with zinc-oxide and eugenol cement.

The histologic analysis, at 21 days, showed that of 17 pulps capped with anorganic bovine dentin, seven were undergoing calcific repair, two pulps were mildly inflamed, but had little reparative dentin formation and eight pulps showed local abscess formation. The marked difference between various pulps capped with the same medicament raised suspicion of bacterial contamination and it was decided to examine the pulps of the second animal (42 days) for bacteria using the Brown and Brenn staining technique.

The 42-day analysis revealed that seven of the 20 experimental pulps were successfully healed. There were four pulps which showed delayed healing and minimal repair and the remaining nine pulps

were severely inflamed with abscess formation in the chambers. When the sections stained for bacteria were examined, a direct correlation was found between delayed healing and the presence of bacteria. Bacteria were found in the pulps of all teeth in which there was no bridging. On the other hand, no bacteria were found in the seven pulps which were successfully repaired.

With the exception of two pulps capped with autogenous dentin chips, all the control teeth healed uneventfully. The two failures had minute abscesses in the exposure areas.

The reparative dentin stimulated by the autogenous and heterogenous cappings was atubular, while that initiated by the calcium hydroxide was occasionally tubular and resembled osteodentin.

The relationship between inflammation and calcific repair was well illustrated in several sections. In one particular section of a pulp capped with autogenous chips, reparative dentin was being laid down on one side of the chip while on the opposite side where slight inflammation existed, no calcification was found. A similar type of situation is illustrated in Figures 9 and 10.

Conclusions

Macacca Speciosa provides an ideal dentition for studies of the dental pulp and the handling procedures described allow the investigator to carry out lengthy operations with minimal concern for animal loss.

In this study, autogenous dentin chips did not interfere with pulp healing, but played a major role in bridge-building.

Since all of the failures in the experimental teeth seem to have been caused by bacterial contamination, anorganic bovine dentin appears to stimulate reparative dentin formation in an exposed pulp. The pulp reaction to the bovine dentin was similar to that of the autogenous dentin. Therefore, it may also be concluded that acid-pepsin hydrolysis is a suitable method for removing the antigenic factor from dentin.

The fact that over half the total sample of pulps in this study were contaminated in spite of the aseptic procedure should be a warning to future investigators to re-examine their techniques and to be prepared for surprises.

The results of this study have shown the importance of staining sections for bacteria before analyzing pulp reactions to a specific treatment. Pulp inflammation which might seem to have been caused by a particular drug or procedure might in fact be due to bacteria.

REFERENCES

1. Rowe, A.H.R.: Molar endodontics. Brit. Dent. J. 121:501, 1966.

2. Spooner, S.: Guide to sound teeth or a popular treatise on the teeth. New York, Waley and Long, 1836.

3. Arthur, R.S.: Treatment of dental caries, complicated with disorders of the pulp and periodontal membrane. Amer. J. Dent. Sci., 3, 1852.

4. Chase, H.S.: Results of pulp treatment. Dent. Cosmos. 8: 517:1866.

5. Hitchcock, T.B.: Oxychloride of zinc to protect dental pulps. Dent. Cosmos. 13:131, 1871.

6. Arnold, O.: Post-mortem treatment of the dental pulp. Dent. Cosmos. 35:126, 1893.

7. Burdell, H.: Observations on the Structure, Physiology, Anatomy and Diseases of the Teeth. New York, Gould and Newman, 1838.

8. Koecker, L.: Principles of Dental Surgery. Baltimore, Amer. Lib. of Dent. Sci., 1842.

9. Allen, W.H.: Post-mortem treatment of the dental pulp. Dent. Cosmos. 7:422, 1866.

10. Atkinson, W.H.: Dental pulps and their treatment. Dent. Cosmos. 10:281, 1868.

11. Christensen, W.E.: Some comments upon "The Herbst Method of Treating Pulps." Dent. Cosmos. 35:363, 1893.

12. Harlan, A.W.: "The Herbst Method of Treating Pulps." Dent. Cosmos. 35:169, 1893.

13. Boennecken, H.: About new methods of treating diseased pulps. Ohio Dent. J. 19:183, 1899.

14. Buckley, J.P.: A rational treatment for putrescent pulps. Dent. Review, 18:1193, 1904.

15. Boennecken, H.: Pulp amputation. Brit. Dent. J. 30:1348, 1909.

16. Rebel, H.H.: Uber die Ausheilung der Freigelegten Pulpa. Dtsch. Zahnheilk Votr. H.55, 1922.

17. Hermann, B.W.: Dentinobliteration der Wurzelkanalenach Behandlung mit Kalzium.Zahnarztl. Rundschau, 21:887, 1930.

18. Teuscher, G.W. and Zander, H.A.: A preliminary report on pulpotomy. Northwestern Univ. Bull. 39:4, 1938.

19. Zander, H.A.: Reaction of the pulp to calcium hydroxide. J. Dent. Res. 18:373, 1939.

20. Pisanti, S. and Sciaky, I.: Origin of calcium in the repair wall after pulp exposure in the dog. J. Dent. Res. 43:641, 1964.

21. Stark, M.M., Myers, H.M., Morris, M. and Gardner, R.: The localization of radioactive Ca^{45} over exposed pulps in rhesus monkey teeth: a preliminary report. J. Oral Ther. 1:290, 1964.

22. Zander, H.A. and Law, D.B.: Pulp management in fractures of young permanent teeth. J. Amer. Dent. Ass. 29:737, 1942.

23. Berk, H.: The effect of calcium hydroxide-methyl cellulose paste on the dental pulp. J. Dent. Child. 17:65, 1952.

24. Gardner, A.F.: Partial pulpectomy, an accepted treatment for primary and young permanent teeth. Oral Surg. 3:498, 1950.

25. Hess, W.: The treatment of teeth with exposed vital pulps. Int. Dent. J. 1:10, 1950.

26. Tannenbaum, N.I.: Pulp capping with zinc oxide-eugenol and calcium hydroxide: clinical studies in 135 patients. J. Dent. Child. 18:16, 3rd quarter, 1951.

27. Chatterton, D.B.: Pulp curettage. J. Amer. Dent. Ass. 45: 462, 1952.

28. Patterson, S.S. and Van Huysen, G.: The treatment of pulp exposures. Oral Surg. 7:194, 1954.

29. Seltzer, S. and Bender, I.B.: The dental pulp. Philadelphia and Montreal, J. B. Lippincott Co., pp. 192-195, 1965.

30. Via, W.F.: Evaluation of deciduous molars treated by pulpotomy and calcium hydroxide. J. Amer. Dent. Ass., 50:34, 1955.

31. Quigley, M.B.: Effect of blood clotting on hamster pulp exposures. Oral Surg. 10:315, 1957.

32. Ostrom, C.A. and Lyon, H.W.: Pulpal response to chemically treated heterogenous bone in pulp capping sites. Oral Surg. 15:362, 1962.

33. Quarterly Cumulative Index Medicus 32:589, 1942.

34. Bower, A.B.: Preserving vitality in pulp exposed teeth. D. Survey 23:1069, 1947.

35. Kutscher, A.M.: Penicillin sodium capping of vital cariously exposed pulps in adults. Dent. Dig. 56:388, 1950.

36. Gilberg, S.L.: Penicillin pulp therapeutics. U.S. Armed Forces Med. J. 2:203, 1951.

37. Rosen, L.J.: Deciduous pulp capping: its present status and a report on penicillin pulp therapeutics. J. Missouri Dent. A. 32:11, 1952.

38. James, J.E., Englander, H.R. and Massler, M.: Histologic response of amputated pulps to calcium compounds and antibiotics. Oral Surg. 10:975, 1957.

39. Seltzer, S. and Bender, I.B.: Some influences affecting repair of the exposed pulps of dogs' teeth. J. Dent. Res. 37:678, 1958.

40. Burke, G.W. and Holmes, T.K.: Effect of local antibacterial agents on bacteria in dental pulps of rats. J. Dent. Res. 41:1105, 1962.

41. Baker, G.R.: Topical antibiotic treatment of infected dental pulps in monkeys. Thesis, Indiana Univ. School of Dent., 1966.

42. Sidky, E.: Combined hydrocortisone-omnacillin-calcium hydroxide therapy in pulpotomy. Egyptian Dent. J. 3:9, 1957.

43. Mager, M.E.: Treatment of pulpitis with synthetic steroids. A review and results of a pilot study. Dent.Pract. 14:505, 1964.

44. Schroeder, A. and Triadon, H.: The pharmacotherapy of pulpitis. Oral. Surg. 15:345, 1962.

45. Olsen, Paul: Clinical experiences with a cartico-antibiotic preparation in conservative treatment of the pulp. J. Canad. Dent. A. 30:771, 1964.

46. Fiore-Donno, G. and Baume, L.J.: Effects of capping compounds containing corticosteroids on the human dental pulp. Helvetica Odont. Acta 6:23, 1962.

47. Baume, L.J.: Clinical and pathohistological aspects of current endodontic therapy. Int. Dent. J. 16:30, 1966.

48. Lawson, B.F. and Mitchell, D.F.: Pharmacologic treatment of painful pulpitis. Oral Surg. 17:47, 1964.

49. Mullaney, T.P., Lawson, B.F. and Mitchell, D.F.: Pharmacologic treatment of pulpitis: A continuing investigation. Oral Surg. 21:497, 1966.

50. Olsen, Paul:Further experience with triamcinolone-demethylchlor-tetracycline for conservative endodontic treatment. J. Canad. Dent. A. 32:522, 1966.

51. Fiore-Donno, G. and Baume, L.J.: Clinical and histological response of human pulp to direct application of triamcinolone-demethylchlortetracycline. J. Canad. Dent. A. 32:527, 1966.

52. Kakehasi, S., Stanley, H.R. and Fitzgerald, R.J.: The effects of surgical exposures of dental pulps in germ-free and conventional laboratory rats. Oral Surg. 20:340, 1965.

53. Stellwagen, T.C.: The natural dentine for capping exposed teeth pulps. Amer. Dent. Ass. Trans. 110-112, 1879.

54. Buckley, J.P.: Protecting the dental pulp and filling the canals of pulpless teeth. Pacific Dent. Gaz. 32:593, 1924.

55. Datwyler, G.: Klinische and histologische Untersuchungen uber einige Pulpauberkappungsmethoden. Schweiz. Monchr. Zahnhilk. 3:291, 1921.

56. Kronfeld, R.: Uber den Ausgang traumatischer Pulpenschadigung. Z. Stomat. 27:846, 1929.

57. Feldmann, G.: Die apikale Paradentitis im Lichte des Experimentes oder der gegenwartige Stand der Probleme der Wurzelbehandlung und Wurzelfullung im Lichte des Experimentes. Verlag Meusser, Berlin, 1931.

58. Feldman, G.L.: Neue Wege in der Therapie von Zahnen mit entzundeter Pulpa. Vjschr. Zahnheilk. 48:211-231, 306-315, 1932.

59. Hellner, E.: Patho-histologische rontgenologische Untersuchungen uber die Pulpaamputation. Z. Stomat. 28:742-774, 855-870, 1930.

60. Neuwirt, F.: Prekryti otevrene drene. Zubni Lekarstvi 28: 1, 1928.

61. Neuwirt, F.: Die reparativen Fahigkeiten der pulpa. Z. Stomat. 31:291, 1933.

62. Pribyl, D.: Die biologische Reaktionsfahigkeit der amputierten nichtentzundeten Pulpa. Rapp. Congr. dent. int. (FDI) (Section III), p. 130, Paris, 1931.

63. Zajfe, M. and Schatzker, K.: Klinische rontgenologische Untersuchungen uber die chirurgischbiologische Methode der Pulpaamputation. Dtsch. zahnarztl. Wschr. 41:390, 1938.

64. Lowenstein, H.: Klinische und rontgenologische Untersuchungen uber drei Methoden der aseptischen Vitalamputation der Pulpa. Zahnarztl. Rdsch. 34:1370, 1934.

65. Hoffman, F.: Die direkte Pulpa-uberkappung nach der Dentinsplittermethode. Schweiz. Mschr. Zahnheilk 47:115, 1937.

66. Castagnola, L.: Die lebenderhaltung der Pulpa in der Konservierenden Zahnheilkunde, Hanser Verlag, Munchen, 1953.

67. Glass, R.L. and Zander, H.A.: Pulp healing. J. Dent. Res. 28:97, 1949.

68. Van Huysen, G. and Boyd, D.A.: Operative procedures and the tooth. J. Prosth. Dent. 3:818, 1953.

69. Kalnins, V. and Frisbie, M.E.: The effect of dentine fragments on the healing of the exposed pulp. Arch. Oral. Biol. 2:96, 1960.

70. Doyle, W.A.: A comparison of the formocresol technique with calcium hydroxide pulpotomy technique. Thesis, Indiana Univ. School of Dentistry, 1961.

71. Spedding, R.H.: The effect of formocresol and calcium hydroxide on the dental pulps of rhesus monkeys. Thesis, Ind. University School of Dent., 1963.

72. Schaffer, E.M.: Cementum and dentin implants in a dog and rhesus monkey. J. Perio. 28:125, 1957.

73. Kuttler, Y.: A precision and biological root canal filling technique. J. Amer. Dent. Ass. 56:38, 1958.

74. Prudden, J.F.: Enhancement of acceleration of wound healing produced by cartilage preparations: with a report on the use of cartilage preparation in clinically chronic ulcers and in primarily closed human surgical incisions. Arch. Surg. 89: 1046, 1964.

75. Prudden, J.F. and Allen, John: The clinical acceleration of healing with a cartilage preparation. J. Amer. Med. A. 192: 352, 1965.

76. King, A. and Orbach, J.: The stump-tailed Macagne: a promising laboratory primate. Science, 139:45, 1963.

77. Brown, J.H. and Brenn, L.: A method for the differential staining of gram-positive and gram negative bacteria in tissue sections. Bull. of John Hopkins Hosp. 48:69, 1931.

78. Norman, R.D., Swartz, M.L., Phillips, R.W.: Studies on film thickness, solubility and marginal leakage of dental cements. J. Dent. Res. 42:950, 1963.

79. Subramian, V.: Response of the amputated dental pulp to topically applied anorganic bone. Brit. Dent. J. 110:221, 1961.

80. Conrado, C.A.: Bacteriostatic and bacteriocidal properties of calcium hydroxide and calcium hydroxide containing cavity bases in vitro using carious dentin as the source of micro-organisms. Thesis, Univ. of Alabama, 1965.

81. Seltzer, S., Bender, I.B., Kaufman, I.J. and Moodnik, R.: Alkaline phosphatase in reparative dentinogenesis. Oral Surg. 15:859, 1962.

82. Johanson, B.I., Persson, I. and Manera, P.: Histologic effects of collagen and chondroitin sulphate as capping agents in amputated rat molar pulps. Arch. Oral Biol. 8:503, 1963.

CURRICULUM VITAE

MARTIN J. WALSHE

October 19, 1940	Born to Alice and Sean Walshe in Bundoran, Ireland
1952 - 1957	De La Salle School Bally Shannon, Ireland
1957 - 1960	University College Galway, Ireland
1960 - 1963	University College Dublin, Ireland
January, 1964	B.D.S., National University of Ireland
February, 1964 - July, 1965	General Practice
1965 - 1967	Graduate student, Indiana University School of Dentistry, Indianapolis, Indiana, U.S.A.
September 8, 1966	Married Fionnuala O'Briain

Professional Societies

American Dental Association
American Society of Dentistry for Children
Irish Dental Association

ABSTRACT

Pulp Reaction to Anorganic Bovine Dentin

by

MARTIN J. WALSHE
1121 West Michigan Street, Indianapolis, Indiana, U.S.A.

A study was made to determine if heterogenous dentin, devoid of its antigenic potential, would stimulate reparative dentinogenesis in the dental pulp. The teeth of two monkeys were capped with bovine dentin mixed with methyl cellulose and histologic analysis was made at 21 and 42 days post-operatively. At the 21-day interval, seven of the 17 teeth capped with the experimental material were successfully repaired with atubular dentin. The remaining 10 teeth showed varying degrees of inflammation and repair. The teeth of the second animal (42 days) were stained for bacteria as an additional diagnostic tool. A direct correlation was found between delayed healing and inflammation and presence of bacteria in the pulp. No bacteria were found in pulps which were successfully repaired. It was concluded that anorganic bovine dentin seemed to induce calcific repair of the dental pulp in the absence of bacteria. Autogenous dentin chips appeared to have the same effect. The importance of including a bacteriologic stain in the histologic analysis of pulp capping studies was demonstrated.